

**REMARKS**

The foregoing amendments amend the specification to reflect the 371 status. In addition, the multiple dependency of the claims has been removed to reduce the PTO filing fee.

Attached hereto is a marked-up version of the changes made to the specification and claims by the current amendment. The attached pages are captioned "**Version with markings to show changes made**".

Favorable action on the merits is solicited.

Respectfully submitted,

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By



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DESCRIPTION

IMMUNOASSAY OF PIVKA-II

*Amg*  
*A1* ~~This application is a 371 of PCT/JP00/03550 filed June 1, 2000.~~  
5 Technical Field:

The present invention relates to an immunoassay through an antigen-antibody reaction for specifically and highly sensitively measuring PIVKA-II (Protein Induced by Vitamin K Absence or Antagonist-II) in serum or plasma by adding to reagents thrombin  
10 and/or an antibody reacting with human fibrin-like related substances.

Background Art:

Along with AFP(  $\alpha$  -fetoprotein), PIVKA-II (Protein  
15 Induced by Vitamin K Absence or Antagonist-II) is measured widely in clinical examination laboratories as a hepatic cell tumor detecting marker which specifically increases in hepatic cell cancer patients. Generally, magnetic beads, glass beads, plastic plates, latexes or the like on which PIVKA-II specific monoclonal  
20 or polyclonal antibodies are adsorbed are subjected to a first reaction with serum or plasma, then after being washed for B/F separation, a second reaction where human prothrombin specific polyclonal or monoclonal antibodies labeled with an enzyme, a fluorescent material, a radioisotope, an Ru complex or the like are  
25 added is carried out, and further after being washed for B/F

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## CLAIMS

1. An immunoassay of PIVKA-II in serum or plasma, comprising the steps of:
  - 5 adding to reagents thrombin and/or an antibody reacting with human fibrin-like related substances, and measuring PIVKA-II in serum or plasma.
2. The immunoassay according to claim 1, wherein a thrombin-containing animal serum and/or purified thrombin are used as the thrombin.
- 10 3. ~~The immunoassay according to claim 1 or 2, wherein an anti-fibrinogen antibody and/or an anti-fibrin antibody are used as the antibody reacting with human fibrin-like related substances.~~

[illegible]